

pET23d:HSVTKII vector is described by Black et al., *Proc. Nat'l Acad. Sci. USA* 93:3525-3529, 1996. Restriction of pET23d:HSVTK-Nar, which is the pET23d:HSVTKII vector with an engineered *NarI* site, by *SacI* and *NarI* allowed removal of TK sequences and replacement by a 1kb *NarI/SacI* fragment from the vector, pLXSN. This vector was designated pET23d:HSVTK-Nar Dummy.--

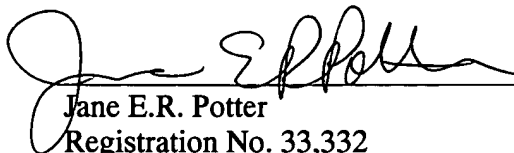
### REMARKS

The enclosed electronic and paper copies of the Sequence Listing include no new material that goes beyond the original application as filed, but is supplied to fulfill the requirements as outlined in the Examiner's communication and Notice to Comply. Furthermore, the above amendments, which merely direct the insertion of the Sequence Listing and insertion of sequence identifiers, include no material that goes beyond the original application as filed. Applicant respectfully submits that the above-identified application is now in compliance with 37 C.F.R. §§ 1.824-1.825.

Attached hereto is a marked-up version of the changes made to the specification and claims by the current amendment. The first of the attached pages is captioned Version with Markings to Show Changes Made."

Respectfully submitted,

Seed Intellectual Property Law Group PLLC

  
Jane E.R. Potter  
Registration No. 33,332

JEP:ljt

Enclosures:

Postcard

PTO Transmittal Form

Computer Diskette

Declaration Regarding Computer Diskette

Paper Copy of Sequence Listing

Copy of Notice to Comply

701 Fifth Avenue, Suite 6300

Seattle, Washington 98104-7092

Phone: (206) 622-4900

Fax: (206) 682-6031

Wpn\240083\429\seq\amendment

Application No. : 09/173,463  
Docket No. : 240083.429  
Examiner : Christian Fronda

**VERSION WITH MARKINGS TO SHOW CHANGES MADE**

**In the Specification:**

Paragraph beginning at page 8, line 11, has been amended as follows:

Figure 3 depicts the nucleic acid and amino sequences of: Wild-type (SEQ ID Nos. 12 & 13), TKF105 (SEQ ID Nos. 55 & 56), TKI208 (SEQ ID Nos. 22 & 23), and TKF2 (SEQ ID Nos. 57 & 58) TK for codons 165 to 175.

Paragraph beginning at page 9, line 6, has been amended as follows:

Figure 14 is an illustration which depicts the nucleotides which were randomized in the LIF-ALL library (SEQ ID NO:47), as well as the results of selection.

Paragraph beginning at page 9, line 8, has been amended as follows:

Figure 15 is a table which shows amino acid substitutions of selected (SEQ ID Nos. 71-88) and unselected clones (SEQ ID Nos. 89-104).

Paragraph beginning at page 9, line 12, has been amended as follows:

Figure 17 is a table which shows nucleotide changes in selected TK mutants (SEQ ID Nos. 59-70).

Paragraph beginning at page 9, line 20, has been amended as follows:

Figure 21 shows semi-randomized oligonucleotides (SEQ ID Nos. 114 & 115) used to generate a second generation of TK mutants having amino acid substitutions in residues 159-161 and 168-169.

Paragraph beginning at page 9, line 23, has been amended as follows:

-- Figure 22 illustrates the use of particular oligonucleotides (SEQ ID Nos. 116-121) to construct TK mutants having amino acid substitutions in residues 112-132.

Paragraph beginning at page 9, line 27, has been amended as follows:

-- Figure 24 illustrates a nucleotide sequence (SEQ ID NO:48) and deduced amino acid sequence (SEQ ID NO:49) representative of a human guanylate kinase.

Paragraph beginning at page 9, line 29, has been amended as follows:

Figure 25 illustrates a nucleotide sequence (SEQ ID NO:50) and deduced amino acid sequence (SEQ ID NO:51) of a representative murine guanylate kinase.

Paragraph beginning at page 84, line 3, has been amended as follows:

Seven mutants that demonstrated required activities were selected for further study. Table VI shows the wild-type TK (SEQ ID NO:105) and deduced amino acid sequence of these seven mutants (SR11, SR26, SR39, SR4, SR15, SR32, SR53; SEQ ID Nos. 106, 107, 108, 109, 110, 111, and 112 respectively). --

Please replace the paragraph beginning at page 87, line 18, with the following rewritten paragraph:

-- To construct a dummy vector for insertion of the random sequences, a *NarI* (or *KasI*) site was introduced into pET23d:HSVTKII by site-directed mutagenesis, using primer DMO1358 (5'-GTCTCGGAGGCGCCAGCACC-3'; SEQ ID NO:113) within the wild-type thymidine kinase open reading frame at nucleotide position 276 from the ATG. The pET23d:HSVTKII vector is described by Black et al., *Proc. Nat'l Acad. Sci. USA* 93:3525-3529, 1996. Restriction of pET23d:HSVTK-Nar, which is the pET23d:HSVTKII vector with an

engineered *NarI* site, by *SacI* and *NarI* allowed removal of TK sequences and replacement by a 1kb *NarI/SacI* fragment from the vector, pLXSN. This vector was designated pET23d:HSVTK-Nar Dummy. --